Exhibit N

INTERNATIONAL STANDARD

ISO 22262-1

First edition
2012-07-01

Air quality — Bulk materials —

Part 1: Sampling and qualitative determination of asbestos in commercial bulk materials

Qualité de l'air — Matériaux solides —

Partie 1: Échantillonnage et dosage qualitatif de l'amiante dans les matériaux solides d'origine commerciale

Reference number
ISO 22262-1:2012(E)

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Published in Switzerland

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Foreword

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International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

ISO 22262-1 was prepared by Technical Committee ISO/TC 146, *Air quality*, Subcommittee SC 3, *Ambient atmospheres*.

ISO 22262 consists of the following parts, under the general title *Air quality — Bulk materials*:

— *Part 1: Sampling and qualitative determination of asbestos in commercial bulk materials*

The following part is under preparation:

— *Part 2: Quantitative determination of asbestos by gravimetric and microscopical methods*

Introduction

In the past, asbestos was used in a wide range of products. Three varieties of asbestos found extensive commercial application. Chrysotile accounted for approximately 95 % of consumption, and this variety is therefore likely to be encountered most frequently during the analysis of samples. Materials containing high proportions of chrysotile asbestos were used in buildings and in industry for fireproofing, thermal insulation, and acoustic insulation. Chrysotile was also used to reinforce materials to improve fracture and bending characteristics. A large proportion of the chrysotile produced was used in asbestos–cement products. These include flat sheets, tiles and corrugated sheets for roofing, pipes and open troughs for the collection of rainwater, as well as pressure pipes for supply of potable water. Chrysotile was also incorporated into products such as decorative coatings and plasters, glues, sealants and resins, floor tiles, gaskets, and road paving. In some products, chrysotile was incorporated to modify rheological properties, e.g. in the manufacture of ceiling tile panels and oil drilling muds. Long textile grade chrysotile fibre was also used to manufacture woven, spun, felted and paper products.

Amosite and crocidolite accounted for almost all of the remaining asbestos use. Amosite was widely used as fireproofing and in thermal insulation products, e.g. pipe coverings and insulating boards. Crocidolite was also used as fireproofing and in thermal insulation products, but was particularly prized because it is highly resistant to acids, flexible enough to be spun and has high tensile strength for reinforcement. Crocidolite found application as a reinforcing fibre in acid containers such as those used for lead–acid batteries, in high-performance textiles and gaskets, and was particularly important for the manufacture of high-pressure asbestos cement pipes for delivery of potable water.

Three other types of asbestos are currently regulated. Materials containing commercial anthophyllite are relatively rare, but they have also been used as a filler and reinforcing fibre in composite materials, and as a filtration medium. Tremolite asbestos and actinolite asbestos were not extensively used commercially, but some occurrences of tremolite asbestos in surfacing materials and fireproofing have been found in Japan. Tremolite asbestos and actinolite asbestos sometimes occur as contaminants of other commercial minerals. Other minerals can also occur as asbestos. For example, richterite asbestos and winchite asbestos occur at mass fractions between 0,1 % and 6 % associated with vermiculite, formerly mined at Libby, Montana, USA. Vermiculite from this source was widely distributed and is often found as loose fill insulation and as a constituent in a range of construction materials and fireproofing.

While the asbestos mass fraction in some products can be very high and in some cases approach 100 %, in other products the mass fractions of asbestos used were significantly lower and often between 1 % and 15 %. In some ceiling tile panels, the mass fraction of asbestos used was close to 1 %. There are only a few known materials in which the asbestos mass fraction used was less than 1 %. Some adhesives, sealing compounds and fillers were manufactured in which asbestos mass fractions were lower than 1 %. There are no known materials in which asbestos was intentionally added at mass fractions lower than 0,1 %.

In this part of ISO 22262, procedures for collection of samples and qualitative analysis of commercial bulk materials for the presence of asbestos are specified. The primary method used to identify asbestos is polarized light microscopy. Because of the wide range of matrix materials into which asbestos was incorporated, polarized light microscopy cannot provide reliable analysis of all types of asbestos-containing materials in untreated samples. The applicability of polarized light microscopy can be extended by the use of simple treatments such as ashing and treatment with acid. Optionally, either scanning electron microscopy or transmission electron microscopy may be used as an alternative or confirmatory method to identify asbestos.

Although this part of ISO 22262 specifies that, optionally, a visual estimate of the asbestos mass fraction within very broad ranges may also be made, it is recognized that the accuracy and reproducibility of such estimates is very limited. Quantitative determination of the asbestos content can be needed for a number of reasons, e.g. assessment and management of the risk from asbestos materials in buildings or to comply with regulatory definitions for asbestos-containing materials. The necessity to quantify asbestos in a material depends on the maximum mass fraction that has been adopted by the jurisdiction to define an asbestos-containing material for the purpose of regulation. Definitions range from “any asbestos” to 0,1 %, 0,5 % or 1 %. For jurisdictions in which an asbestos-containing material is defined as one containing “any asbestos”, a particular problem is how to determine whether a material does not contain asbestos, since all methods have limits of detection.

For practical purposes, since no known commercial materials exist in which commercial asbestos was intentionally added at mass fractions lower than 0,1 %, this part of ISO 22262 specifies that samples be classified as asbestos-containing (i.e. containing more than 0,1 % asbestos) if either chrysotile, amosite, crocidolite or anthophyllite, or any of these varieties in combination, is detected in the analysis. When the definition of an asbestos-containing material is either 0,5 % or 1 %, depending on the nature of the product, it is often necessary to proceed to other parts of this International Standard in order to quantify the asbestos for the purpose of defining the regulatory status of the material.

The occurrence of tremolite, actinolite or richterite/winchite in a material is usually a consequence of natural contamination of the constituents, and the detection of these minerals does not necessarily indicate that the mass fraction is more than 0,1 % asbestos. Accordingly, determination of the regulatory status of these materials by any of the criteria can often be achieved only by quantitative analysis. Since these minerals were not specifically mined and utilized for their fibrous properties, they may also occur in materials as either non-asbestiform or asbestiform analogues, or as mixtures of both. Evaluation of these types of material may require a more detailed analysis.

Simple analytical procedures such as polarized light microscopy are not capable of detecting or reliably identifying asbestos in some types of commercial products containing asbestos, either because the fibres are below the resolution of optical microscopy or because the matrix material adheres too strongly to the fibres. For these types of product, it may be necessary to utilize electron microscopy.

For a list of parts of this International Standard, see the Foreword.

The method specified in this part of ISO 22262 is based on MDHS 77,[11] VDI 3866 Part 1,[13] VDI 3866 Part 4,[14], VDI 3866 Part 5,[15], AS 4964-2004,[8] EPA/600/R-93/116,[10] and NF X46-020:2008.[12]

Air quality — Bulk materials — Part 1: Sampling and qualitative determination of asbestos in commercial bulk materials

IMPORTANT — The electronic file of this document contains colours which are considered to be useful for the correct understanding of the document. Users should therefore consider printing this document using a colour printer.

1 Scope

This part of ISO 22262 specifies methods for sampling bulk materials and identification of asbestos in commercial bulk materials. This part of ISO 22262 specifies appropriate sample preparation procedures and describes in detail the procedure for identification of asbestos by polarized light microscopy and dispersion staining.

This part of ISO 22262 also specifies simple procedures for separation of asbestos fibres from matrix materials such as asphalt, cement, and plastics products. Optionally, identification of asbestos can be carried out using scanning electron microscopy or transmission electron microscopy with energy dispersive X-ray analysis. Information is also provided on common analytical problems, interferences and other types of fibre that may be encountered in the analysis.

This part of ISO 22262 is applicable to qualitative identification of asbestos in specific types of manufactured asbestos-containing products and commercial minerals. This part of ISO 22262 is applicable to the analysis of fireproofing, thermal insulation, and other manufactured products or minerals in which asbestos fibres can readily be separated from matrix materials for identification.

NOTE This part of ISO 22262 is intended for use by microscopists who are familiar with polarized light microscopy methods and the other analytical procedures specified (References [16]–[19]). It is not the intention of this part of ISO 22262 to provide instruction in the fundamental analytical techniques.

2 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

2.1
achromat
microscope objective in which chromatic aberration is corrected for two wavelengths and spherical aberration and other aperture-dependent defects are minimized for one other wavelength (usually about 550 nm)

EXAMPLE One wavelength less than about 500 nm, the other greater than about 600 nm.

NOTE This term does not imply any degree of correction for curvature of image field; coma and astigmatism are minimized for wavelengths within the achromatic range.

[ISO 10934-1:2002,[3] 2.6]

2.2
acicular
shape shown by an extremely slender crystal with cross-sectional dimensions which are small relative to its length, i.e. needle-like

[ISO 13794:1999,[4] 2.1]

2.3
alpha refractive index
α
lowest refractive index exhibited by a fibre

2.4
amphibole
group of rock-forming ferromagnesium silicate minerals, closely related in crystal form and composition, and having the nominal formula:

$$A_{0-1}B_2C_5T_8O_{22}(OH,F,Cl)_2$$

where

A is K, Na

B is Fe^{2+}, Mn, Mg, Ca, Na

C is Al, Cr, Ti, Fe^{3+}, Mg, Fe^{2+}

T is Si, Al, Cr, Fe^{3+}, Ti

NOTE In some varieties of amphibole, these elements can be partially substituted by Li, Pb, or Zn. Amphibole is characterized by a cross-linked double chain of Si-O tetrahedra with a silicon:oxygen ratio of 4:11, by columnar or fibrous prismatic crystals and by good prismatic cleavage in two directions parallel to the crystal faces and intersecting at angles of about 56° and 124°.

[ISO 13794:1999,[4] 2.2]

2.5
amphibole asbestos
amphibole in an asbestiform habit

[ISO 13794:1999,[4] 2.3]

2.6
analyser
polar used after the object to determine optical effects produced by the object on the light, polarized or otherwise, with which it is illuminated

NOTE It is usually positioned between the objective and the primary image plane.

[ISO 10934-1:2002,[3] 2.117.1]

2.7
anisotropy
state or quality of having different properties along different axes

EXAMPLE An anisotropic transparent particle can show different refractive indices with the vibration direction of incident light.

2.8
asbestiform
specific type of mineral fibrosity in which the fibres and fibrils possess high tensile strength and flexibility

[ISO 13794:1999,[4] 2.6]

2.9
asbestos
term applied to a group of silicate minerals belonging to the serpentine and amphibole groups which have crystallized in the asbestiform habit, causing them to be easily separated into long, thin, flexible, strong fibres when crushed or processed

NOTE 1 The Chemical Abstracts Service Registry Numbers of the *most common* asbestos varieties are: chrysotile (12001-29-5), crocidolite (12001-28-4), grunerite asbestos (amosite) (12172-73-5), anthophyllite asbestos (77536-67-5), tremolite asbestos (77536-68-6) and actinolite asbestos (77536-66-4).

[ISO 13794:1999,[4] 2.7]

NOTE 2 Other varieties of asbestiform amphibole, such as richterite asbestos and winchite asbestos (Reference [20]), are also found in some products such as vermiculite and talc.

2.10
aspect ratio
ratio of length to width of a particle

[ISO 13794:1999,[4] 2.10]

2.11
Bertrand lens
intermediate lens which transfers an image of the back focal plane of the objective into the primary image plane

NOTE The Bertrand lens is used for conoscopic observation in polarized light microscopy and for adjustment of the microscope illuminating system, especially in phase-contrast and modulation-contrast microscopy.

[ISO 10934-1:2002,[3] 2.87.2]

2.12
birefringence
quantitative expression of the maximum difference in refractive index due to double refraction

[ISO 10934-1:2002,[3] 2.16]

2.13
camera length
equivalent projection length between the specimen and its electron diffraction pattern, in the absence of lens action

[ISO 13794:1999,[4] 2.12]

2.14
chrysotile
fibrous mineral of the serpentine group which has the nominal composition:

$Mg_3Si_2O_5(OH)_4$

NOTE Most natural chrysotile deviates little from this nominal composition. In some varieties of chrysotile, minor substitution of silicon by Al^{3+} may occur. Minor substitution of magnesium by Al^{3+}, Fe^{2+}, Fe^{3+}, Ni^{2+}, Mn^{2+} and Co^{2+} may also be present. Chrysotile is the most prevalent type of asbestos.

[ISO 13794:1999,[4] 2.13]

2.15
cleavage
breaking of a mineral along one of its crystallographic directions

[ISO 13794:1999,[4] 2.14]

2.16
cleavage fragment
fragment of a crystal that is bounded by cleavage faces

NOTE Crushing of non-asbestiform amphibole generally yields elongated fragments that conform to the definition of a fibre, but rarely have aspect ratios exceeding 30:1.

2.17
crossed polars
state in which the polarization directions of the polars (polarizer and analyser) are mutually perpendicular

[ISO 10934-1:2002,[3] 2.117.2]

2.18
***d*-spacing**
distance between identical adjacent and parallel planes of atoms in a crystal

[ISO 13794:1999,[4] 2.18]

2.19
dispersion
variation of refractive index with wavelength of light

[ISO 7348:1992,[1] 05.03.26]

2.20
dispersion staining
effect produced when a transparent object is immersed in a surrounding medium, the refractive index of which is equal to that of the object at a wavelength in the visible range, but which has a significantly higher optical dispersion than the object

NOTE Only the light refracted at the edges of the object is imaged, and this gives rise to colours at the interface between the object and the surrounding medium. The particular colour is a measure of the wavelength at which the refractive index of the object and that of the medium are equal.

2.21
electron diffraction
technique in electron microscopy by which the crystal structure of a specimen is examined

[ISO 13794:1999,[4] 2.19]

2.22
electron scattering power
extent to which a thin layer of substance scatters impinging electrons from their original directions

[ISO 13794:1999,[4] 2.20]

2.23
energy dispersive X-ray analysis
EDXA
measurement of the energies and intensities of X-rays by use of a solid-state detector and multichannel analyser system

[ISO 13794:1999,[4] 2.22]

2.24
eucentric
condition in which the area of interest of an object is placed on a tilting axis, at the intersection of the electron beam with that axis, and is in the plane of focus

[ISO 13794:1999,[4] 2.23]

2.25
extinction
condition in which an optically anisotropic object appears dark when observed between crossed polars

[ISO 10934-1:2002,[3] 2.51]

NOTE Extinction occurs when the vibration directions of the crystal are parallel to the vibration directions in the polarizer and analyser.

2.26
extinction angle
angle between the extinction position and the position at which the length of a fibre is parallel to the polarizer or analyser vibration directions

2.27
fibril
single fibre of asbestos which cannot be further separated longitudinally into smaller components without losing its fibrous properties or appearances

[ISO 13794:1999,[4] 2.25]

2.28
fibre
elongated particle which has parallel or stepped sides

[ISO 13794:1999,[4] 2.26]

NOTE For the purposes of this part of ISO 22262, a fibre is defined to have an aspect ratio greater than or equal to 3:1.

2.29
fibre bundle
structure composed of parallel, smaller diameter fibres attached along their lengths

NOTE A fibre bundle may exhibit diverging fibres at one or both ends.

[ISO 13794:1999,[4] 2.27]

2.30
gamma refractive index
γ
highest refractive index exhibited by a fibre

2.31
habit
characteristic crystal growth form, or combination of these forms, of a mineral, including characteristic irregularities

[ISO 13794:1999,[4] 2.30]

2.32
high-efficiency particulate air filter
HEPA
filter that is at least 99,97 % efficient by volume on 0,3 µm particles

[ISO 14952-1:2003,[6] 2.13]

2.33
isotropic
having the same properties in all directions

[ISO 14686:2003,[5] 2.23]

2.34
Köhler illumination
method of illuminating specimens in which an image of the illumination source is projected by a collector into the plane of the aperture diaphragm in the front focal plane of the condenser, which then projects an image of an illuminated field diaphragm at the opening of the collector into the specimen plane

2.35
lamda zero
λ_0
matching wavelength corresponding to the dispersion staining colour shown by a particle in an immersion medium

NOTE At this wavelength, the particle and the immersion medium have the same refractive index.

2.36
matrix
material in a laboratory sample within which fibres are dispersed

2.37
Miller index
set of either three or four integer numbers used to specify the orientation of a crystallographic plane in relation to the crystal axes

[ISO 13794:1999,[4] 2.33]

2.38
pleochroism
property of an optically anisotropic medium by which it exhibits different brightness and/or colour for different directions of light propagation, or for different vibrations, on account of variation in selective spectral absorption of transmitted light

2.39
polarized light
light in which the vibrations are partially or completely suppressed in certain directions at any given instant

NOTE The vector of vibration may describe a linear, circular or elliptical shape.

[ISO 10934-1:2002,[3] 2.88.1]

2.40
polarizer
polar placed in the light path before the object

[ISO 10934-1:2002,[3] 2.117.4]

2.41
polar
device which selects plane-polarized light from natural light

[ISO 10934-1:2002,[3] 2.117]

2.42
refractive index
n
ratio of the speed of light (more exactly, the phase velocity) in a vacuum to that in a given medium

[ISO 10934-1:2002,[3] 2.124]

2.43
retardation
difference in optical path length expressed in wavelengths, length units or phase angles between two mutually perpendicular plane-polarized waves

[ISO 10934-1:2002,[3] 2.128]

2.44
selected area electron diffraction
technique in electron microscopy in which the crystal structure of a small area of a sample is examined

[ISO 13794:1999,[4] 2.38]

2.45
serpentine
group of common rock-forming minerals having the nominal formula:

$Mg_3Si_2O_5(OH)_4$

[ISO 13794:1999,[4] 2.39]

2.46
sign of elongation
description of the directions of the high and low refractive indices in a fibre

NOTE The fibre is described as positive when the higher refractive index is parallel to the length of the fibre, and negative when the lower refractive index is parallel to the length of the fibre.

2.47
temperature coefficient of refractive index
measure of the change of refractive index of a substance with temperature

2.48
twinning
occurrence of crystals of the same species joined together at a particular mutual orientation, and such that the relative orientations are related by a definite law

[ISO 13794:1999,[4] 2.41]

2.49
unopened fibre
large diameter asbestos fibre bundle that has not been separated into its constituent fibrils or fibres

[ISO 13794:1999,[4] 2.42]

2.50
zone-axis
line or crystallographic direction through the centre of a crystal which is parallel to the intersection edges of the crystal faces defining the crystal zone

[ISO 13794:1999,[4] 2.43]

3 Symbols and abbreviated terms

$\frac{dn}{dT}$	change of RI of an immersion medium per degree Celsius change of temperature
n_D^{25}	RI of a liquid for the sodium D line (589,3 nm) and at a temperature of 25 °C
α	lowest RI of an anisotropic particle
β	intermediate RI of an anisotropic particle
γ	highest RI of an anisotropic particle
λ_0	wavelength at which the RI of a particle is equal to the RI of the liquid in which it is immersed
ED	electron diffraction
EDXA	energy dispersive X-ray analysis
FWHM	full width, half maximum

HEPA	high-efficiency particle absolute
MEC	mixed esters of cellulose
PC	polycarbonate
PCOM	phase contrast optical microscopy
PLM	polarized light microscopy
RI	refractive index
SAED	selected area electron diffraction
SEM	scanning electron microscopy
TEM	transmission electron microscopy

4 Principle

4.1 General

A suitable tool is used, in compliance with the relevant safety regulations, to take a sample from the material to be analysed. The sample is then appropriately packed and labelled for transportation to the laboratory.

A representative sample of the bulk material is initially examined using a stereo-binocular microscope. Typical fibres are removed using tweezers and mounted in appropriate liquid immersion media on slides for examination by polarized light microscopy. Asbestos fibres are identified based on morphology, colour, pleochroism, and the α (lowest) and γ (highest) refractive indices qualitatively assessed using the dispersion staining technique. Detection of commercial asbestos (chrysotile, amosite, crocidolite or anthophyllite), either alone or in combination, is assumed to indicate that the asbestos is present at a mass fraction exceeding 0,1 %. Optionally, a visual estimate of the asbestos mass fraction is reported in one of several broad mass fraction ranges. Tremolite, actinolite and richterite/winchite are identified by the same procedure, but since they are usually present as contaminants of mineral products, detection of these minerals does not provide information as to their minimum mass fraction. Optionally, fibres may be identified by SEM or TEM.

4.2 Substance determination

This International Standard specifies a number of reference methods for determination of asbestos in solid materials. This part of ISO 22262 provides a method for qualitative analysis of specific commercial products for the presence of asbestos (chrysotile, amosite, crocidolite, tremolite, actinolite, anthophyllite and richterite/winchite). Other parts of this International Standard provide methods for the analysis of specific types of commercial products for which the use of PLM on the untreated sample yields unacceptable rates of error, and for the quantification of asbestos in the low mass fraction range below approximately 5 %.

4.3 Type of sample

The method specified in this part of ISO 22262 is applicable to sampling and analysis of commercial products from which individual fibres of asbestos can be manually separated from the matrix material, either by picking fibres from surfaces and newly fractured surfaces, or after chemical treatments, acid extraction or ashing, such that the fibres can be identified by one of the specified identification methods. This part of ISO 22262 is generally applicable to asbestos-containing building materials such as fireproofing, thermal pipe and boiler insulations, asbestos cement, plasters, roofing, and other similar materials. The method is also applicable to the identification of asbestos in a range of other industrial minerals and materials.

4.4 Range

Experience from proficiency testing has shown that the range of this part of ISO 22262, when it is applied to a suitably prepared sample in which the asbestos fibres are sufficiently large to be optically visible using a low-

magnification stereomicroscope, is from less than 0,1 % to 100 %. The lower end of the range can be extended downwards by use of appropriate techniques.

4.5 Limit of detection

The limit of detection of this method is defined as the detection and identification of one fibre or fibre bundle in the amount of sample examined. The limit of detection that can be achieved depends on:

a) the nature of the matrix of the sample;

b) the size of the asbestos fibres and bundles;

c) the use of appropriate sample preparation and matrix reduction procedures;

d) the amount of time expended on examination of the sample;

e) the method of analysis used — PLM, SEM or TEM.

With appropriate matrix reduction procedures that are tailored to the nature of the sample, the limit of detection can be significantly lower than 0,01 %.

4.6 Limitations of PLM in the detection of asbestos

The ability to detect and identify asbestos by PLM is limited by the resolution of the optical microscope and sometimes by the masking effects of other materials that comprise the balance of the sample. Asbestos fibres with widths below approximately 0,2 µm are unlikely to be detected by PLM. However, for all varieties of amphibole asbestos, and most varieties of chrysotile, a large proportion of the mass comprises fibres that exceed this width and, because of this, asbestos can be reliably detected by PLM. Accordingly, provided that the nature of the matrix material on the microscope preparation is such that it does not obscure any asbestos fibres that might be present, a non-detected result by PLM indicates that the mass fraction of asbestos is below the limit of detection.

One commercial source of chrysotile presents problems of detection by PLM. Chrysotile originating from the Coalinga deposit in California, USA, contains no fibrils longer than approximately 30 µm and, if these are well dispersed in a sample matrix, the majority of the chrysotile is below the size that can be reliably detected and identified by PLM. The range of application of Coalinga chrysotile is limited to floor tiles, ceiling tiles, drywall joint compounds, mastics, paints, sealants, adhesives, drilling mud, moulded cement building products, and as filler in some plastics. There is a high probability that this variety of chrysotile may not be detected by PLM, even when present in high mass fractions. The size distribution of Coalinga chrysotile makes it unsuitable for most other applications in which asbestos was used and the possibility that it will be encountered in other types of product can generally be discounted. If, on the basis of PLM examination, Coalinga chrysotile is suspected to be present, it is recommended that the sample be examined by electron microscopy.

Asbestos fibres may not be detected by PLM because they are obscured by the matrix of the sample. The matrix reduction methods specified in this part of ISO 22262 are intended to minimize the possibility of failing to detect asbestos in such samples.

5 Sample collection

5.1 Requirements

5.1.1 Sampling apparatus. Depending on the nature of the material to be sampled, an appropriate tool is required for collection of the sample. If the material is soft, such as thermal insulation or fireproofing, a knife or scalpel may be sufficient. In other situations, a cork borer may be used to sample all of the layers of a layered material. If the material is hard, e.g. asbestos–cement, tools such as pliers, a wire cutter, hammer and chisel or rotating hole saw can be needed.

5.1.2 HEPA vacuum cleaner. A HEPA vacuum cleaner, approved for asbestos, is required for cleaning around the sampling location after collection of the sample to minimize dispersion of asbestos-containing dust or particulate matter.

5.1.3 Materials and supplies for sampling.

5.1.3.1 Wetting agent. A wetting agent may be used to limit the generation of airborne dust during the collection of the sample. Water, or water to which a small amount of surfactant has been added, may be applied to the surface before sampling using a spray bottle or brush.

IMPORTANT If a sample is being collected for the purpose of product identification, use no wetting agent, since this may result in alteration of the sample composition by addition of surfactant, and by dissolution and loss of water-soluble constituents.

5.1.3.2 Filler. After collection of the sample, a minor repair may be necessary to seal the damaged area. Depending on the circumstances, spray paint, touch-up paint or plaster may be used.

5.1.3.3 Sample containers. Appropriate dust-tight containers are required for packaging the sample. Plastic bags with "zip" closures or bottles with screw caps may be used.

5.1.3.4 Labels. A method for labelling samples is required. Self-adhesive paper labels may be used. Alternatively, a waterproof marker may be sufficient for field use.

5.1.3.5 Dust mask. A dust mask with filter approved for respiratory protection against airborne asbestos fibres. Approved filters conform to either the National Institute for Occupational Safety and Health (NIOSH) P100 or the European Standard EN 143[9] P3 specification. Other types of personal protective equipment may be used if warranted by the situation.

5.1.3.6 Light. Either a flashlight or an appropriate light source is required for collection of samples in dark locations.

5.1.3.7 Plastic bags. Labelled plastic bags of appropriate size that can be closed tightly and are required to collect the waste generated during sampling. Bags containing waste should be placed inside another tightly closed plastic bag.

5.1.3.8 Cleaning supplies. Cleaning materials, such as disposable paper towels and a supply of water, are required for cleaning sampling tools to avoid cross-contamination between samples.

5.1.3.9 Location identifiers. The use of some means of identifying the precise location from which each sample is taken is recommended, since it may be necessary to resample the material at a later date to resolve discrepancies if they arise. A location identifier is invaluable if the sample collected is found not to be representative of the overall area, such as if the sample has been taken from a patch in a location that has been repaired. A specific colour of spray paint, or appropriate permanent labels applied to the precise location, may be used.

5.2 Procedure

5.2.1 Safety precautions

Handling asbestos is regulated by many jurisdictions, and regulations often specify a variety of procedures to ensure that individuals performing work and those in close proximity are not exposed to excessive concentrations of airborne asbestos. Exceptions from the regulations are generally permitted for some types of activity that are minimally invasive, such as the removal of material samples for analysis.

IMPORTANT — Care is necessary during sampling of materials that may contain asbestos, and precautions should be taken to avoid creating and inhaling airborne asbestos particles when sampling materials suspected of containing asbestos. If the handling instructions in this clause are followed, it may be

assumed that the level of dust meets the thresholds of safety defined in the regulations. In exceptional cases, more extensive precautions may be necessary to prevent the release of airborne fibres.

Sometimes different materials may have been applied to a surface as several layers. It is recommended that samples of all of the individual layers be collected. If a borer or hole-sawing device is used to penetrate several layers, the device should be operated so that it rotates slowly. This ensures that only coarse turnings are produced. High-speed devices are not recommended, since it is then necessary to take more complex safety precautions such as local suction and filtration to collect the dust generated.

5.2.2 Sample size requirements

5.2.2.1 General

Although only a few milligrams of sample are required for the analytical methods specified, it is necessary to take into account the homogeneity of the material, and to ensure that the sample is of sufficient size to be representative of the material under investigation. If inspection shows that the material is finely divided and homogeneous when examined visually, or if the nature of the material is recognized as such from previous knowledge, a minimum sample size of approximately 1 cm^3 generally provides sufficient material for analysis. However, a minimum volume of 10 cm^3 is recommended for materials such as sprayed fireproofing, and as much as 1 000 cm^3 for materials such as loose-fill vermiculite.

5.2.2.2 Representative sample

A wide range of asbestos-containing materials was used in the past. Experience is very valuable in the selection of the materials to be sampled and sampling can be facilitated by the use of all available prior knowledge about the materials or components from which the sample is being collected. It is essential that the sample collected be representative of the composition of the product with respect to its asbestos content. Although many asbestos-containing materials may seem to be homogeneous when visually examined, they can be quite inhomogeneous in the microscopic size range. This is particularly the case for materials such as texture coats, in which the fragments of aggregate are significantly larger than the other constituents of the material.

In some types of material, particularly those that have been mixed at a building site, rather than a commercial product manufactured and mixed under a formulation and quality control procedure, the asbestos may not be distributed homogeneously within the material. For these types of materials, it is necessary to collect a larger sample to ensure that the sample is representative of the material.

It is recommended that a portion of the sample be archived, because further examination of the sample is often the only way in which potential questions can be resolved.

In addition to the problem of inhomogeneity, the possibility that repairs using materials from different sources may have occurred needs to be considered. For example, during renovation or repairs, some asbestos-free ceiling tiles may have been installed in a suspended ceiling, the balance of which contain asbestos, for no other reason than such ceiling tiles were readily available at the time. During repairs or rebuilding, other materials of the same appearance, but having different compositions, may have been used to repair damage to fireproofing, thermal insulation or bulkheads.

It is important to recognize that the analytical result relates only to the actual sample tested. If the sample collected is not representative, the result will not be representative of the material.

Annex A, which lists the asbestos-containing materials most frequently used, provides guidance for identifying different types of material.

5.2.2.3 Number of samples

The number of samples to be taken is dependent on the nature of the material, whether the material is homogeneous or inhomogeneous, and the size of the area under consideration. In the case of materials known from prior experience to be homogeneous, it may be sufficient to collect one sample, although collection of more than one sample provides additional confidence that the results are representative of the material being sampled. When materials are suspected to be inhomogeneous, it is necessary to collect several samples and

to ensure that each of the samples is of sufficient size. If it is intended to determine the range of asbestos content in an area of material, it is necessary to analyse all of the samples individually. Otherwise, such samples may be combined before analysis in order to ensure that the sample analysed represents the mean asbestos mass fraction of the material.

5.2.2.4 Precautions to avoid cross-contamination between samples

It is most important that precautions be taken to ensure that cross-contamination of samples does not occur. Clean all tools used for collecting samples prior to initial use and again after collection of each sample. Use a new and unused container or plastic bag for each sample, and double-bag each sample.

5.2.2.5 Sampling strategy

Selection of the sampling locations depends on the type of area being sampled and on the nature of the product suspected to contain asbestos.

The selection of the sampling locations shall be made in accordance with any national regulations.

The material being sampled may be known to be homogeneous, e.g. a manufactured packing material or sheet material. Samples should be collected at locations that are as inconspicuous as possible. Locations that exhibit prior superficial damage or locations behind readily detached covers are particularly suitable, provided that there are no reasons to suspect that the material in such locations is not representative.

IMPORTANT — Ensure that the sampling location is not at a position where repair using a different material has previously occurred.

If the material under test has a layered structure, e.g. in the case of multilayer pipe insulations or multilayer floor coverings, include all layers of the material in the collected sample. Include any coverings or adhesive layers, such as coatings or glues. Do not attempt to separate the layers under field conditions; separation of individual layers for analysis is best performed under controlled conditions in the laboratory.

If the product under test is behind a wall cladding or other covering, power sockets or light switch recesses are frequently suitable as locations for collection of material samples. If it is not possible to gain access in this manner, it is necessary to cut the claddings or coverings open in order to enable sample collection. These openings should be made at a location that detracts from the visual appearance as little as possible, e.g. behind baseboards.

5.2.2.6 Taking the samples

Release of airborne asbestos fibres from asbestos-containing materials may occur before or during the sampling. The use of containment measures may be necessary. If the material is such that a significant release of airborne asbestos fibres may occur during collection of the sample, sample carefully and moisten the sampling location with water from a spray bottle, a water-soaked brush or a moist paper towel. A moist paper towel is also useful to clean contaminated surfaces after the sample has been collected.

Water should not be used if samples are being collected in the vicinity of operating electrical equipment.

a) For many types of homogeneous material, it is usually possible to collect small amounts of sample without visibly defacing the material and without incurring any significant release of airborne fibres.

b) If the material appears to be homogeneous, collect a sample area more than 1 cm^2 in the case of thin materials, or a volume greater than 1 cm^3 in the case of materials having a thickness of several centimetres. Remove the sample by breaking it off with pincers or preferably using a sharp cutting tool. If the material appears to be inhomogeneous, collect a sufficient amount of sample to give confidence that the volume of sample is representative of the material.

c) Place each sample in an individual dust-tight container.

d) Wipe the sampling site and the immediate surroundings, keeping them moist, or clean the area around the sample location using a vacuum cleaner with a HEPA filter.

e) If necessary, seal the exposed surface from which the sample was taken using touch-up paint, glue or other appropriate sealant.

f) Affix, if applicable and agreed to by the facility administration, a permanent identification marker to the exact location from which the sample is removed.

5.2.2.7 Sample labelling

Label the sample container clearly, either by using a permanent marker pen or by attaching a permanent adhesive label. Confirm that the sample label corresponds to the information on any identification marker affixed to the sampling location.

5.2.2.8 Sampling record

Make a record of the sample that contains at least the following information:

a) full description of the type of material;

EXAMPLE Thermal insulation, board, floor tile.

b) all details recorded on the sample label;

c) precise description of the sampling location;

d) building identification;

e) identification of the room (if applicable);

f) location in the room from which the sample was collected;

g) the date that the sample was collected;

h) the name of the person who collected the sample;

i) whether the sample is a composite derived from the combination of separately collected samples;

j) whether the sample is a multilayer sample — for multilayer samples, the positions of each of the relevant layers shall be noted.

If the sampling location is not adequately specified by the details specified in a) to f), then, in addition:

k) make a sketch or take a photograph (record the number of the photograph); or, record the position from which the sample was taken on a plan of the building (the drawing identification shall also be noted in the record);

l) report any other relevant data that are available with respect to the sample.

An example of a suitable sampling record is shown in Annex G.

5.2.2.9 Chain of custody

If there is any possibility that the results of sampling and analysis will be subject to litigation or legal scrutiny, it is most important that records be made of all transfers of samples between individuals, starting with the individual who collected the samples through to acceptance of the samples by the analyst. A chain of custody form shall be used for this purpose, on which the date of each transfer and the name of each individual who has relinquished or accepted possession of the samples are recorded.

5.2.2.10 Storage and transport

The samples shall be packaged in dust-tight containers (double if necessary) and a label shall be affixed to the package of samples, indicating that they may contain asbestos. Take care to ensure that unauthorized persons do not have access to the samples. There are no special requirements with respect to climate conditions

for storage and transport of the samples. After the samples have been analysed, they shall be archived for whatever period of time is specified by the individual submitting them to the analytical laboratory.

6 Sample preparation

6.1 General

It is sometimes not possible to identify asbestos in bulk materials because of interference by other constituents, either because the mass fraction of asbestos is too low or because the asbestos is so inhomogeneously distributed that a large amount of the sample would need to be examined in order to reliably detect the asbestos that is present. In these cases, various chemical or physical preparation methods can be used prior to the microscopic examination to remove a large proportion of the non-asbestos constituents, thus facilitating the detection of asbestos in the smaller amount of material that remains.

6.2 Removal of organic materials by ashing

Chrysotile is often difficult to detect when mixed with large amounts of cellulose, or if it is well dispersed in organic matrices such as asphalt or poly(vinyl chloride) (PVC). Also, some other organic fibres such as spider webs and wool have optical properties similar to those of chrysotile. Ashing of the sample at a temperature of 485 °C for a period of approximately 10 h removes the organic constituents with very little effect on the optical properties of chrysotile. Although the colour and optical properties of amosite and crocidolite are altered by this oxidation treatment as a consequence of conversion of some ferrous iron [Fe(II)] to ferric iron [Fe(III)], many of the fibres can often still be identified by PLM. The optical properties of tremolite, actinolite, anthophyllite and richterite/winchite are almost unaffected by this treatment. The heat treatment does not otherwise affect the composition of any of the asbestos varieties, and they can all be identified by electron microscopy after the treatment.

6.3 Removal of soluble constituents by acid treatment

Matrix constituents such as calcite and gypsum often coat asbestos fibres so that their optical properties cannot be reliably examined. These constituents also often constitute a large proportion of the sample mass. Stirring of a sample in 2 mol/l hydrochloric acid for approximately 15 min removes many matrix constituents, and this improves the ability to identify and quantify asbestos. The acid treatment slightly reduces the refractive indices of chrysotile, and it is necessary to account for this when identifying chrysotile by PLM. Do not heat chrysotile in acid at temperatures exceeding 60 °C. This acid treatment does not affect the optical properties of any of the other asbestos varieties.

6.4 Sedimentation and flotation

Some materials contain large sizes of aggregate or sand that can be separated in water suspension by sedimentation or flotation. A large proportion of constituents such as vermiculite or perlite can be separated by flotation. Sand or small solid aggregate sediment in water much more rapidly than most of the asbestos, and in some samples a large proportion of the sand or aggregate can be separated from the fraction that contains any asbestos.

6.5 Combination of gravimetric reduction procedures

The procedures specified in 6.2, 6.3 and 6.4 may be combined as appropriate for the particular sample.

It is generally recommended that the procedures be used sequentially in the order given.

7 Analysis by PLM

7.1 Requirements

7.1.1 Stereo-binocular microscope, for initial observation of samples. The examination is facilitated if the microscope has a continuous range of magnification from approximately 10× to 40×.

7.1.2 Polarized light microscope, capable of Köhler (or Köhler-type) illumination is needed for fibre identification. The following optical accessories are necessary:

a) light source with blue “daylight” filter;

b) focusing sub-stage condenser with a numerical aperture (NA) greater than or equal to that of the objective in use, with a field-limiting adjustable aperture;

c) focusing ocular with magnification of 10 times or 12 times, with a cross-hair graticule;

d) strain-free objectives with magnifications of 4 times, 10 times, and 40 times or similar magnifications;

e) polarizer and removable analyser, the vibration directions of which can be adjusted such that they are at 90° to each other, and can be aligned with the cross-hair in the focusing ocular;

f) slot between the polarizer and analyser to allow accessory plates to be inserted at an angle of 45° to the polarizer and analyser vibration directions;

g) removable retardation plate with approximately 530 nm retardation, with known slow and fast vibration directions;

h) dispersion staining objective with magnification of 10 times or 40 times, or a demonstrated functional equivalent (MDHS 77[11]);

i) Bertrand lens or a focusing telescopic ocular to allow observation of the back focal plane of the objective lens;

j) level rotating specimen stage for which the centre of rotation can be centred relative to the optical axis of the microscope for each of the objective lenses.

7.1.3 Dust extract hood. Handling and manipulation of bulk materials suspected to contain asbestos shall be performed in a suitable dust extract hood, so that neither the analyst nor the laboratory environment is exposed to airborne asbestos fibres.

7.1.4 Sample preparation.

7.1.4.1 Refractive index liquids. The majority of commercial asbestos-containing products contain only chrysotile, amosite or crocidolite, or mixtures of these three types of asbestos. Identification of these three types of asbestos can be achieved using liquids of RI 1,550, 1,680 and 1,700. The RI values of these liquids are specified for light of wavelength 589,3 nm (sodium D line) at a temperature of 25 °C.

For identification of tremolite, actinolite, anthophyllite and richterite/winchite, RI liquids in the range 1,605 to 1,660 are required, at intervals of 0,005.

Suitable calibrated RI liquids are commercially available, and a set of liquids with RIs from 1,500 to 1,700, at intervals of 0,005, gives sufficient range and discrimination.

If commercially available RI liquids cannot be obtained, a set of liquids sufficient for use in this part of ISO 22262 can be prepared (References [16][21]) using common chemical reagents as specified in Table 1.

Table 1 — Reagents for preparation of RI immersion media

Reagent	n_D^{25}	$\frac{dn}{dT}$
Glycerol triacetate	1,427 7	–0,000 48
Ethyl cinnamate	1,557 4	–0,000 48
Bromobenzene	1,557 0	–0,000 54
Iodobenzene	1,617 3	–0,000 54
1-Chloronaphthalene	1,630 4	–0,000 44
1-Bromonaphthalene	1,658 0	–0,000 45
1-Iodonaphthalene	1,700 4	–0,000 44
Diiodomethane	1,739 0	–0,000 70
Commercially available RI media, and the reagents listed here, should be used in accordance with applicable safety precautions.		

Table 2 shows the mixtures of reagents required to prepare a set of RI immersion media. The three primary RI liquids for identification of chrysotile, amosite and crocidolite are indicated in Table 2 in bold type (1,550, 1,680 and 1,700). Tremolite, actinolite or anthophyllite can often be identified using only RI liquids 1,605 and 1,630, also indicated in Table 2 in bold type. Tremolite, actinolite or anthophyllite may be encountered in which the refractive indices are high because of increased iron mass fraction, and use of other RI liquids in Table 2 may be necessary in order to assess the refractive indices.

Table 2 — Mixtures and single compounds required for RI liquids

Liquid n_D^{25}	Liquid 1	Volume fraction, liquid 1 %	Liquid 2	Volume fraction, liquid 2 %	$\frac{dn}{dT}$
1,545	Ethyl cinnamate	90,44	Glycerol triacetate	9,56	–0,000 48
1,550	**Ethyl cinnamate**	**94,30**	**Glycerol triacetate**	**5,70**	**–0,000 48**
1,555	Ethyl cinnamate	98,15	Glycerol triacetate	1,85	–0,000 48
1,560	Bromobenzene	95,03	Iodobenzene	4,97	–0,000 54
1,605	**Iodobenzene**	**79,60**	**Bromobenzene**	**20,40**	**–0,000 54**
1,610	Iodobenzene	87,89	Bromobenzene	12,11	–0,000 54
1,615	Iodobenzene	96,19	Bromobenzene	3,81	–0,000 54
1,620	1-Chloronaphthalene	85,83	Bromobenzene	14,17	–0,000 45
1,625	1-Chloronaphthalene	92,64	Bromobenzene	7,36	–0,000 45
1,630	**1-Chloronaphthalene**	**100**	—	—	**–0,000 44**
1,635	1-Bromonaphthalene	78,99	Bromobenzene	21,01	–0,000 47
1,640	1-Bromonaphthalene	84,05	Bromobenzene	15,95	–0,000 46
1,645	1-Bromonaphthalene	89,11	Bromobenzene	10,89	–0,000 46
1,650	1-Bromonaphthalene	94,18	Bromobenzene	5,82	–0,000 46
1,655	1-Bromonaphthalene	99,24	Bromobenzene	0,76	–0,000 45
1,660	1-Bromonaphthalene	90,48	1-Iodonaphthalene	9,52	–0,000 45
1,680	**1-Iodonaphthalene**	**54,31**	**1-Bromonaphthalene**	**45,69**	**–0,000 44**
1,700	**1-Iodonaphthalene**	**100**	—	—	**–0,000 44**

7.1.4.2 Asbestos reference standards. Asbestos reference standards are required. Suitable sets of standards are SRM 1866[1)] (chrysotile, crocidolite and amosite) and SRM 1867[1)] (tremolite, actinolite and anthophyllite) from the US National Institute of Standards and Technology (NIST), see Table 3, or from the UK Health and Safety Executive (HSE) [Chrysotile (Canada and Zimbabwe), crocidolite, amosite, tremolite, actinolite and anthophyllite][2)] see Table 4. SRM 1867 tremolite and actinolite are particularly useful for qualitative discrimination between tremolite and actinolite. The International Mineralogical Association (IMA) (References [23][24]) has specified that values of the mass fraction ratio Mg/(Mg + Fe) below 0,9 are defined as tremolite, and those above 0,9 are defined as actinolite. SRM 1867 tremolite has a value of 0,84, and SRM 1867 actinolite has a value of 0,94, providing reference samples representing compositions just below and just above the IMA boundary. It is important to recognize that the IMA boundary between tremolite and actinolite is only a convention within a continuum of composition in which the iron and magnesium mass fractions vary in a reciprocal manner.

Table 3 — Optical properties of SRM 1866 and SRM 1867 reference asbestos samples

Property	Chrysotile	Amosite	Crocidolite	Anthophyllite	Tremolite	Actinolite
Colour	White	Grey–brown	Blue	Light brown	White	White
Pleochroism	None	Very weak	α: Blue, γ: grey	None	None	None
Birefringence	Low	Medium	Low	Medium	Medium	Medium
Sign of elongation	Positive	Positive	Negative	Positive	Positive	Positive
Extinction	Parallel	Parallel	Parallel	Parallel	16,6°	15,9°
γ	1,556	1,701	—[a]	1,636	1,634	1,639
α	1,549	1,679	—[a]	1,615	1,606	1,613

[a] For crocidolite, the certificate of analysis states: "Because strong absorption in the visible light range results in anomalous dispersion characteristics that would not be useful to the analyst, no certified values of refractive index are reported for riebeckite".

Table 4 — Optical properties of HSE reference asbestos samples

Property	Chrysotile (Canada)	Chrysotile (Zimbabwe)	Amosite	Crocidolite	Anthophyllite	Tremolite	Actinolite
Colour	White	White	Grey–brown	Blue	White	White	Pale green
Pleochroism	None	None	Very weak	α: Blue, γ: grey	None	None	γ-Green, α: grey
Birefringence	Low	Low	Medium	Low	Medium	Medium	Medium
Sign of elongation	Positive	Positive	Positive	Negative	Positive	Positive	Positive
Extinction	Parallel	Parallel	Parallel	Parallel	Parallel	Parallel	Parallel
γ	1,552	1,552	1,692	1,696	1,624	1,632	1,652
α	1,544	1,544	1,676	1,688	1,608	1,616	1,644

NOTE The data for the HSE reference asbestos samples notes that, "as with all natural minerals, the reference samples may contain traces of other minerals. In particular, the anthophyllite asbestos sample contains a fibrous variety of talc which may be distinguished by its ribbon-like morphology and generally lower refractive indices".

For those laboratories that are unable to obtain either the NIST or the HSE reference asbestos samples, the Union Internationale Contre le Cancer (UICC) standard reference samples of asbestos (Reference [25]) may be used, see Table 5. These samples were widely distributed internationally, and can still be obtained.

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1) Example of a suitable product available commercially from the US National Institute of Standards and Technology (NIST). This information is given for the convenience of users of this document and does not constitute an endorsement by ISO of these products.

2) Example of a suitable product available commercially from the from the UK Health and Safety Executive (HSE). See Reference [22]. This information is given for the convenience of users of this document and does not constitute an endorsement by ISO of these products.

However, since the UICC samples were prepared for use in animal studies, they were milled to very small fibre sizes. Also, the UICC samples do not include either tremolite or actinolite.

Table 5 — Optical properties of UICC reference asbestos samples

Property	Chrysotile (Canada)	Chrysotile (Zimbabwe)	Amosite	Crocidolite	Anthophyllite
Colour	White	White	Grey–brown	Blue	White
Pleochroism	None	None	Very weak	α: Blue, γ: grey	None
Birefringence	Low	Low	Medium	Low	Medium
Sign of elongation	Positive	Positive	Positive	Negative	Positive
Extinction	Parallel	Parallel	Parallel	Parallel	Parallel
γ	1,545–1,560	1,553	1,701	1,702	1,620
α	1,545–1,557	1,546	1,679	1,694	1,605

NOTE 1 A range of refractive indices is quoted for the UICC Canadian chrysotile sample. This sample was prepared by blending chrysotile from a number of different mines. Fibres with refractive indices within the approximate ranges specified are present, with birefringence ($\gamma - \alpha$) approximately 0,01.

NOTE 2 The anthophyllite sample also contains a fibrous variety of talc.

7.1.4.3 Sample comminution equipment. An agate mortar and pestle is required for grinding samples to suitable sizes for PLM examination.

7.1.4.4 Microscope slides, 75 mm × 25 mm.

7.1.4.5 Microscope cover glasses, 22 mm × 22 mm. Match the thickness of the cover glasses with that specified by the objective lenses. A thickness of 0,17 mm is required by many commercial objectives.

7.1.4.6 Thermometer, required to measure the temperature of the microscope slide preparation during observation if accurate refractive indices of asbestos fibres are to be recorded.

7.1.4.7 Alcohol or gas burner. A laboratory burner is sometimes useful for discriminating between organic fibres and asbestos fibres.

7.1.4.8 General laboratory supplies. The following supplies and equipment, or equivalent, are required:

a) glassine paper sheets, approximately 15 cm × 15 cm, for examination of samples;

b) scalpel holder and replacement disposable scalpel blades;

c) sampling utensils, including tweezers, needles and spatulas;

d) distilled water;

e) concentrated hydrochloric acid, reagent grade;

f) crucibles, silica or glazed porcelain, with lids;

g) Petri dishes;

h) disposable pipettes;

i) glass filtration assembly, 25 mm or 47 mm diameter;

j) polycarbonate filters, 0,4 µm pore size, 25 mm or 47 mm diameter.

7.1.4.9 Muffle furnace (optional). For ashing of samples to remove interfering organic constituents, a muffle furnace with a temperature range up to 500 °C and a temperature stability of ±10 °C is recommended.

7.1.4.10 Magnetic stirrer (optional). For removal of acid-soluble interfering constituents, a magnetic stirrer with a glass or plastic-coated magnetic stir bar.

7.2 Qualitative analysis by PLM

7.2.1 Calibration

It is essential that the optical components of the PLM be fully understood by the analyst and that the analyst be familiar with the alignment procedure. The alignment of the PLM shall be confirmed prior to conducting any analyses. The designs of microscopes vary and the alignment instructions provided by the manufacturer should be followed. The critical aspects of the alignment are listed in a) to e).

a) The illumination source and sub-stage condenser shall be adjusted so that the field-limiting aperture is in focus (Köhler or Köhler-like illumination).

b) The centre of rotation of the specimen stage shall be aligned with the optical axis of the PLM for each of the objective lenses. This is necessary so that a particle at the centre of the field of view remains at the centre of the field of view during rotation of the stage. This condition is often achieved by centring the rotation for one objective lens, and then laterally adjusting the position of each of the other objective lenses to align their axes with the centre of the stage rotation.

c) The vibration directions of the polarizer and analyser shall be at 90° to each other.

d) The vibration directions of the polarizer and analyser shall accurately coincide with the directions of the cross-hair in the ocular. This can be accomplished using a well-formed birefringent crystal with a known zero extinction angle. Alternatively, orientation plates consisting of an accurately mounted crystal with a fiducial line are commercially available. If the microscope has eyepieces that can be freely rotated, fix the position of the eyepiece containing the cross-hair using adhesive tape, for example.

e) If a mechanical stage is installed on the rotating stage, the directions of the mechanical stage should be adjusted such that the zero angular position of the rotation stage corresponds to lateral motions of the mechanical stage parallel to the polarizer and analyser directions.

On the initial set-up of the PLM, the vibration direction of the polarizer and the orientation of the vibration directions of the 530 nm retardation plate shall be determined. The vibration direction of the polarizer can be determined by examination of a slide preparation of crocidolite with the polarizer in position and the analyser withdrawn. Under these conditions, the direction of the length of the crocidolite fibres when the dark blue pleochroism is displayed is the vibration direction of the polarizer. The orientation of the vibration directions of the 530 nm retardation plate can be determined by examination of a fibre of a known reference material such as amosite or chrysotile, and observing the change of interference colour when the retardation plate is inserted. The slow vibration direction of chrysotile or amosite is parallel to the length of the fibre. If the retardation plate adds to the retardation caused by the fibre, the slow vibration directions of the fibre and the retardation plate are parallel. An interference colour chart is provided in Annex B.

Before using RI liquids for the identification of asbestos, even if certified liquids are purchased, it is recommended that the refractive indices of liquids be confirmed using reference glass samples or a refractometer. If kept tightly capped, the refractive indices of these liquids remain stable for at least 2 years. Some RI liquids degrade when exposed to light, therefore they should be stored in dark bottles, preferably in a dark place.

7.2.2 Sample preparation

For many samples, including fireproofing, thermal insulation and asbestos cement products, fibres that can be removed with tweezers are visible during stereomicroscope examination. Mount typical suspected asbestos fibres on a microscope slide and add a drop of the RI liquid appropriate for the suspected asbestos variety. If the suspected asbestos variety cannot be confirmed using the appropriate RI liquid, mount additional fibres from the sample on slides using RI liquids appropriate for the other asbestos varieties.

7.2.3 Sample analysis

7.2.3.1 Analytical sequence

The analytical techniques described have been shown to give reliable and reproducible results. Alternative methods can be used if their equivalence in terms of detection and identification can be demonstrated. Identification of the asbestos fibres should be based on the following analytical sequence:

a) make a preliminary visual examination of the whole of the laboratory sample to assess the sample type and the required sample treatment (if any) — where possible, take a representative test portion at this stage for direct examination by PLM;

b) carry out any required sample treatment to release or isolate fibres;

c) perform a detailed and thorough search under the stereomicroscope to classify the suspected fibre types present;

d) mount representative fibres in appropriate RI liquids on microscope slides;

e) identify the different fibrous components using PLM.

If no asbestos is detected by these procedures, prepare additional slides using random test portions of a few milligrams and search for thin asbestos fibres using PLM.

7.2.3.2 Preliminary examination

Examine the entire sample visually to describe the type of material or product present, and to establish whether there are visible fibres. Note the nature of any matrix materials, as this may indicate the type of treatment required for the sample. Examine the sample using the stereomicroscope. So far as possible, make an initial determination of the number of fibre types present. Record the appearance, colour and texture of the sample and any fibre types observed. For inhomogeneous or layered samples, it may be necessary to describe each separate layer or part of the sample. Sample preparation and the analysis of the sample are dependent on the quality of the initial visual examination. Also, adequate description of the appearance of the sample is important in establishing whether asbestos is present, or in which part of the sample asbestos is present.

7.2.3.3 Sample treatment

The purpose of any initial treatment of laboratory samples is to release fibres from any matrix and to remove fine particles adhering to the fibres (both of which obscure the optical effects and hinder the identification). It is necessary to break non-friable samples (with tools if necessary) and then to examine newly fractured edges using the stereomicroscope to observe any protruding fibres. If samples contain large pieces of hard materials, grinding the sample may be necessary. Surfaces and edges of hard materials may be abraded to release fibres for examination. Routine procedures used for sample treatment should be fully documented. Any deviations from these procedures for particular samples should be recorded.

Dilute acetic acid or cold dilute hydrochloric acid may be used to remove calcium carbonate (limestone), calcium sulfate (gypsum), and calcium silicate, which are commonly used as binders (e.g. for insulation and asbestos boards) and fillers (e.g. in floor tiles). The removal of calcium magnesium carbonate (dolomite) requires the use of cold concentrated hydrochloric acid. Sufficient acid should be added in small aliquots for several minutes or until effervescence stops. Fibre release may be aided by stirring or by ultrasonic treatment. The sample is then filtered and repeatedly washed with water. Residual acid may degrade the fibres and affect the optical properties, and small crystals of salts may form. The sample may be rinsed with ethanol or other volatile solvents to reduce the drying time.

Organic matrices such as plastics, asphalt, resins or rubber products may require prolonged treatment in solvents to remove the matrix. An effective solvent for any particular sample type can be established only by individual testing or by foreknowledge of the type of matrix. Organic matrices may be removed by treatment in a muffle furnace at 485 °C. However, heating may modify the optical properties of some of the asbestos fibres.

7.2.3.4 Stereomicroscope examination

The original samples or portions of sample that have undergone sample treatment should be examined using the stereomicroscope. For many asbestos-containing materials, asbestos fibres can be detected at magnifications within the range of the stereomicroscope. For other types of asbestos-containing material, it may not be possible to detect asbestos fibres using the stereomicroscope. The aim is to detect small fibre bundles, or individual fibres, and tentatively to assign fibre types based on their appearance. This is usually achieved by placing the sample on a piece of glassine paper or in a suitable container and carrying out a detailed search of the entire sample using needles or tweezers to separate the different fibrous components from the matrix. The appearance of these fibres is then noted. The care and vigilance with which the sample is examined at this stage are important in detecting trace quantities of asbestos. Representative fibres or fibre bundles are then selected and mounted for PLM examination.

Describe layered samples by their appearance, and note each distinct layer as a separate entity. Regulations in some jurisdications require that distinct layers be analysed and reported separately. Other types of inhomogeneous sample will require detailed visual examination of all the different phases observed.

Asbestos is generally recognized by the fineness of its fibres, which are most often present as closely packed bundles of fibrils that will divide along their length when pressure is exerted on them with a probe or tweezers. An analyst will rapidly become familiar with characteristics such as distinctive surface lustre, flexibility, and tensile strength. Initial tentative identification of suspected asbestos fibres at this stage will be confirmed or refuted by subsequent examination using PLM, SEM or TEM.

7.2.3.5 Preparation of samples for PLM examination

A tentative identification based on the stereomicroscope evaluation is used to select the most appropriate RI mounting liquid. Fibres selected shall be dry and relatively free from other particulate matter. Representative fibres or fibre bundles are chosen and are placed on a clean microscope slide into a drop of RI liquid, and a clean cover glass is lowered gently onto the slide, avoiding trapping of air bubbles. The RI of the liquid selected should be 1,550 for suspected chrysotile, 1,680 for suspected amosite, 1,700 for suspected crocidolite, 1,605 for suspected tremolite or anthophyllite, and 1,630 for suspected actinolite or richterite/winchite.

If no fibres have been seen in the bulk sample using the stereomicroscope, or no asbestos fibres have been identified by PLM, then tweezers or probes should be used to take random test portions, after the laboratory sample has undergone suitable treatment (if necessary). At least two microscope slide preparations should be made with appropriate RI liquids for examination by PLM. Any large agglomerates should be teased apart with tweezers or needles, or sheared gently between two microscope slides, to give an even distribution of particles. Selection of large particles or fibre bundles may cause tilting of the cover slip and should be avoided. The amount of sample distributed should be such that the appearance and properties of individual fibres are not obscured by other particles.

7.2.3.6 Identification of asbestos by PLM and dispersion staining

Identification of a single asbestos fibre requires the observation of the following properties in the stated observation modes:

a) morphology — observed in all illumination conditions;

b) colour and pleochroism — observed in plane polarized light;

c) birefringence — observed with crossed polars;

d) extinction characteristics — observed with crossed polars;

NOTE The extinction characteristics can also be observed with crossed polars and a 530 nm retardation plate inserted. Under these conditions, when the interference colour of the fibre matches the background colour, the fibre is at the extinction position.

e) sign of elongation — observed with crossed polars and a 530 nm retardation plate inserted;

f) refractive indices — assessed using a dispersion staining objective with polarizer only inserted.